(19) Europäisches Patentamt
European Patent Office
Office européen des brevets

(11) Publication number: **0 114 686**
B1

(12) # EUROPEAN PATENT SPECIFICATION

(45) Date of publication of patent specification: **04.10.89**

(51) Int. Cl.⁴: **G 01 N 1/28**

(21) Application number: **84100588.7**

(22) Date of filing: **20.01.84**

(54) **Liquid sample handling system.**

(30) Priority: **21.01.83 US 459973**
05.05.83 US 489866

(43) Date of publication of application:
01.08.84 Bulletin 84/31

(45) Publication of the grant of the patent:
04.10.89 Bulletin 89/40

(84) Designated Contracting States:
AT BE CH DE FR GB IT LI NL SE

(56) References cited:
GB-A-1 497 012
US-A-3 188 181
US-A-3 650 306
US-A-4 215 092
US-A-4 422 151

(73) Proprietor: **CETUS CORPORATION**
1400 Fifty-Third Street
Emeryville California 94608 (US)

(72) Inventor: **Salomaa, Kari**
P.O. Box 117
Jenner California (US)
Inventor: **Merrill, Roy**
51 Esta Bueno
Orinda California (US)
Inventor: **Leath, Richard**
941 Elm Street
El Cerrito California (US)
Inventor: **Wennberg, Timothy**
541 Arch Street
San Francisco California (US)
Inventor: **Widunas, Joseph**
202 Clipper Street No. 3
San Francisco California (US)

(74) Representative: **Vossius & Partner**
Siebertstrasse 4 P.O. Box 86 07 67
D-8000 München 86 (DE)

EP 0 114 686 B1

Courier Press, Leamington Spa, England.

Description

The present invention is directed to apparatus and method for handling small liquid samples according to the preambles of claims 1 and 12. Such apparatus and method are disclosed in US—A—4 215 092. More specifically, it is directed to apparatus and method, such as are required in initial filling and serial dilution of liquid samples in microtiter trays where each receptacle holds only about one tenth to ten milliliters of liquid. Such a serial dilution system basically involves mixing the sample with successively increasing proportions of a diluent in separate receptacles thereby to obtain a series of successively decreasing concentrations of the sample. The various sample concentrations can then be assayed to determine a particular property. For example, the sample might be a serum and the assay might be used to determine which concentration of the serum provides optimum results when reacted with a particular substance.

Initially, serial dilution of a sample was performed manually, wherein different proportions of the sample would be mixed with diluent contained in different respective test tubes, for example with the aid of a syringe or pipette. This procedure consumed a considerable amount of time when a number of different concentrations were required. Consequently, machines for automatically or semi-automatically performing serial dilutions were developed. One example of such a machine is disclosed in U.S. Patent US—A—3,188,181. The serial dilution machine disclosed in that patent includes a horizontally movable carriage that accommodates a rack of test tubes. A vertically movable syringe holder housing a plurality of off-the-shelf syringes respectively connected to pipettes is raised and lowered by means of a cam to bring the pipettes into the liquid volume in a row of test tubes within the rack. A cam operated pumping head oscillates the syringes to mix fluid in the pipettes with that in the test tubes. After mixing, fluid is withdrawn from the test tubes, the syringe holder is lifted, and the carriage is incremented to another row of test tubes to repeat the operation. When the serial dilution operation is complete, a first row of test tubes in the rack might contain undiluted concentrations of the sample, the second row of test tubes would contain a 50/50 concentration of the sample, the third row would be a 25/75 concentration, and so forth, depending on the amount of liquid transferred by the pipettes.

The US—A—4,215,092 describes a multichannel pipette having an arrangement of replaceable tip containers in which the individual tip containers are connected together by flexible connecting members which are deformable to permit connection of the tip containers to differing configurations of tip cones.

The US—A—3,650,306 relates to a laboratory dispensing apparatus including a base, a liquid reservoir and well assembly mounted on the base, and a microtitration plate carrier device mounted on the base for reciprocating movement between a forward retracted position and a rearward operative position overlying a portion of the well. A vertically movable pipette dispensing assembly is positioned above the base in operative alignment with the portion of the liquid well so that with the carrier in the forward retracted position the plural pipettes may withdraw microquantities of liquid from the well and then dispense the same into the microtitration plate wells when the carrier is moved to the rearward position. The plural pipettes are supported and a contained in a head unit which is readily removable from the dispensing assembly to permit quick changing of heat units for cleaning purposes.

It is an object of the present invention to provide a novel apparatus and method for automatically effecting transfer of liquid between receptacles without cross contamination or errors in the quantity of liquid so transferred and which is particularly useful in performing serial dilution of small liquid samples with substantially improved performance over that possible with the machine of the previously noted patent. More particularly, when handling certain types of solutions, it is desirable to prevent cross-contamination between various sample concentrations. Typically, this is done in the manual method by utilizing disposable tips on the pipettes that withdraw the liquid from one receptacle and mix it with diluent in another. After each mixing operation, the tip is removed from the pipette and replaced with a clean one. However, heretofore known serial dilution machines do not offer the ability to change tips between each cycle in the serial dilution process. Consequently, when prevention of cross-contamination is desired, the machine must effect a multi-step washing process for the pipettes, or provide a flushing process between each cycle in the process. The washing process substantially increases the time that is required to effect a serial dilution since a number of steps are added in each cycle of the process. The flushing procedure generates a considerable amount of waste liquid that must be disposed of.

Accordingly, it is a more particular object of the present invention to provide a novel automatic serial diluter arrangement that is capable of automatically changing disposable tips on pipettes in a quick and efficient manner in any desired cycle of the serial dilution process from initial fill of the liquid receptacles through any of a selectable plurality of addition or subtraction steps to transfer liquid between the receptacles.

These objects are achieved by the features of the independent apparatus and method claims 1 and 12, respectively.

In a preferred form, at least one work station accommodates a titer tray or similar such structure having plural rows of receptacles for housing the liquid sample and the diluents, and at least a second work station accommodates a rack that houses plural rows of disposable pipette tips. The pipettes themselves have tapered ends that can be inserted into and frictionally engage the tips when a head assembly on which they are mounted is moved downwardly to bring them together. Once the tips are picked up, the table is translated to bring a selected row of wells in the titer tray underneath the tips, and the head is then lowered

to insert the tips within the wells. Some of the sample in the wells is aspirated into the pipettes through actuation of plungers in each pipette and, after raising the head, the table is incremented one or more steps to bring another, generally the next successive, row of wells into registry with the tips. The head is then lowered to insert the tips into the diluent in these wells and the plunger oscillated to mix the sample with the diluent. The head is then raised so that the pipette tip is just above the meniscus of liquid in the well after all the liquid is expelled from the tip. The plunger is then extended to expel all of the liquid with the aid of some excess air from the pipettes and surface tension on any liquid between the tip and the well. The table is then translated to bring the rack housing the tips beneath the pipettes, and a solenoid controlled tip ejector means is actuated to push the tips from the tapered ends of the pipettes and back into the rack. Alternatively, the tips may be ejected through a slot in the table (or at another station on the table) into a trough or collector container. The table then can be incremented one step to bring a fresh set of tips into registry with the pipettes, and the cycle repeated. In a preferred form the volume of each tip is greater than the liquid volume of the titer tray receptacles so that all fluid is moved in and out of the receptacle without substantial liquid contact with the plunger of the pipette.

According to a preferred embodiment of the present invention, a detector can be provided to determine when the pipettes fail to pick up all of the tips in a row, to thereby prevent the situation in which a sample in one well is not serially diluted into all the various desired concentrations through failure to draw the sample into a pipette because of a missing tip. Similarly, a detector to determine that all tips have been ejected from their respective pipettes prior to pick up of the next row of tips may be provided.

The movement of all of the various elements of the machine can be monitored and controlled as desired by a computer, to thereby provide continuous monitoring of the process and flexibility in filling and transferring liquids for the serial dilution process.

The present invention is discussed in greater detail hereinafter with reference to a preferred embodiment thereof illustrated in the accompanying drawings.

Fig. 1 is a perspective view of a serial dilution machine implementing the features of the present invention;

Fig. 2 is a sectional side view of the serial dilution machine taken in the direction of arrows 2—2 in Fig. 1;

Fig. 3 is an enlarged, sectional side view of one embodiment of a pipette tip in a tip support tray well;

Fig. 4 is an enlarged, sectional side view of a preferred form of pipette tip supported in a tip tray well;

Fig. 5 is an enlarged, sectional side view of the plunger and pipette assembly;

Fig. 6 is a top view of the table showing the trays arranged thereon for a 12 × 7 diluter configuration;

Fig. 7 is a top view of the table showing the trays arranged for an 8 × 11 diluter configuration;

Fig. 8 is a sectional front view of the automatic serial dilution machine taken in the direction of arrows 8—8 in Fig. 1;

Fig. 9 is a partial top plan view of the machine;

Fig. 10 is a sectional top view taken along the section line 10—10 of Fig. 8;

Fig. 11 is a sectional top view taken along the section line 11—11 of Fig. 8.

Fig. 12 is a perspective view of an alternate embodiment of a serial dilution machine including a fluid transfer or supply tray between the tip supply tray and the microtiter tray;

Fig. 13 is top plan view of the Fig. 12 table embodiment; and

Fig. 14 is a sectional side view taken in the direction of arrows 14—14 in Fig. 13.

Referring to Figures 1 and 2, an automatic serial dilution machine suitable for carrying out the method of the present invention includes two main movable parts, a horizontally translatable table 10 and a vertically translatable head assembly 12. As best illustrated in Fig. 2, the table 10 is mounted for horizontal translation on hardened guide rods 14 by means of slide bearings 16. Translation of the table is provided by a stepper motor 18 through a pinion 20 connected to the motor and a rack 22 mounted on the underside of the table. Similarly, the head 12 is mounted for vertical translation on guide rods 24 by means of slide bearings 26. Translation of the head assembly is provided by a stepper motor 28 through a pinion 30 and a rack 32.

The head assembly 12 supports a pipette and plunger assembly 34. This assembly includes a series of pipettes 36 or syringes that are arranged in a row transverse to the axis of translation of the table 10. The pipettes are removably attached to the head assembly by means of a mounting block 37, and connecting pins 33 and move therewith. A plunger mechanism 38 is mounted on the head assembly for vertical movement relative to the pipettes. The plunger mechanism includes a series of plunger rods 40, one being disposed respectively within each pipette 36. All of the rods are mounted on a common actuator bar 42 for concurrent vertical movement. The bar 42 is translated along guide rods 44 by means of a stepper motor 46 and a lead screw drive mechanism 48. As best illustrated in the detailed sectional diagram of Fig. 5, translation of the plunger rods 40 relative to the pipettes 36 changes the internal volumes of the pipettes, causing fluid to be aspirated into or expelled from them. An air tight seal is provided between each rod and the top of its associated pipette by means of an O-ring 49, held by grommet 47 and compliance spring 45. Each pipette 36 includes a piston section 39 which is reciprocably mounted in a cylinder 35 formed in mounting block 37. Pipette 36 is thereby restrained vertically by spring 45 so that during the tip loading step, pipette 36 can slide vertically in block 37 against compliance spring 45. This allows all pipettes to

reliably pick up tips of slightly different dimensions and to assure that the open ends of tips 62 are at the same elevation relative to table 10 and titer tray 54.

The table 10 includes two work stations 50 and 52 for respectively accommodating two trays. One of the trays can be a conventional titer tray 54 that includes a matrix arrangement of wells for housing the liquid sample and the diluent. The other tray 56 at the rear work station 52 can be a tip tray that contains a similar arrangement of receptacles that accommodate disposable pipette tips. A typical titer tray contains 96 wells in a 12 × 8 matrix pattern. As illustrated in Fig. 6, the tray 54 can be accommodated at the forward work station 50 in a transverse orientation to perform a 12 × 7 serial dilution, wherein the first row of wells is filled with a predetermined volume of the sample to be diluted, and the remaining wells are filled with the diluent. In this case, the tip tray 56 is also oriented to present twelve receptacles in a row across the width of the tray. Alternatively, as illustrated in Fig. 7, the trays 54 and 56 can be oriented in the longitudinal direction of the table 10 to effect an 8 × 11 serial dilution.

Referring again to the detailed side view of Fig. 5, the bottom end 60 of each pipette 36 is tapered on its exterior surface so as to receive and frictionally engage the inner surface of disposable pipette tip 62 constructed in accordance with the present invention. For example, the tip 62 might be made of a non-wettable polypropylene material. The tips 62 in a row of wells or receptacles 63 in the tip tray 56 are inserted onto and engage the respective endsd of the pipettes 36 when the head assembly 12 is lowered by the stepper motor 28 after the table 10 has brought one row of tips 62 into registry with the pipettes. As indicated, the volume of each tip 62 is a substantial portion of the total volume of the cylinder formed by the barrel of pipette 36 and the interior volume of the tip. As best seen in Figs. 4 and 5, support of each tip 62 in receptacle 63 of tray 56 is either by end support as in Fig. 4 or on ends of the bottom flutes 65 formed on the exterior of tips 62. The wall of receptacles 63 are arranged to center tip 62 for engagement with tapered end 60 of pipette 36.

The subsequent removal of the tips 62 from the pipettes is accomplished with a tip ejector means. The tip ejector means includes a comb-like plate 64 that is best illustrated in Fig. 11. The plate has recesses that accommodate the pipettes, and its teeth surround a substantial portion, e.g., 180°, of the exterior circumference of each pipette barrel. The plate 64 is connected to and supported by a pair of vertically translatable rods 66 mounted on the head assembly 12. These rods are translated by means of a pair of solenoids 68 mounted on the top of the head assembly. When the solenoids 68 are deactuated, the ejector plate 64 is maintained in the upper position illustrated in Fig. 5. Actuation of the solenoids moves the plate vertically downward, to push the tips 62 down and release them from their frictional engagement with the ends of the pipettes 36.

The operation of each of the stepper motors 18, 28 and 46, and the solenoids 68 is controlled by a suitable microprocessor 70. Basically, the microprocessor 70 functions as a pulse generator to control the sequence of operations of each of these elements, and thus the interrelated movements of the table 10, the head assembly 12, the plunger assembly 34 and the tip ejector plate 64 to effect serial dilution of a sample in the tray 54 at the forward work station 50. Since the stepper motors provide a predetermined amount of rotation in response to each actuating pulse applied thereto, accurate positioning of the movable elements can be obtained through appropriate control of the number of actuating pulses supplied by the microprocessor.

In addition to controlling these various movable elements, the microprocessor 70 also monitors their movement through appropriately positioned sensors. For example, a sensor arrangement for the table 10 can include a blade 72 that is attached to and extends from the side of the table, and a Hall-effect sensor 74 that detects when the blade 72, and hence the table 10, passes through a predetermined reference point in its translation. Each time the table passes through this point, the Hall-effect sensor 74 sends a signal to the microprocessor 70 that enables the microprocessor to update information relating to the table's position. Thus, if the stepper motor 18 should miss an actuating pulse during translation of the table, or if the pulse count stored within the microprocessor 70 should not coincide with the position of the table, the error will not be carried over to successive cycles of operation.

In addition to the reference sensor 74, a pair of limit sensors 76 can be disposed at the respective ends of the path of travel of the table. A signal sent by these sensors indicates that the table is nearing the end of its travel, and provides an indication to the microprocessor 70 to interrupt the supply of power to the stepper motor 18 or take some other such corrective action. Similar sensor arrangements are provided to monitor the movement of the head assembly 12 and the plunger bar 42.

Furthermore, a sensor can be provided on the machine to detect whether all of the tips in a row of the tray 56 have been picked up by the pipette assembly. Referring to Figure 8, this sensor can include an electrical-optical mechanism comprising an LED 89 or similar such light emitting device on one side of the table and a photoelectric element 90 on the other side of the table. The two elements are aligned with the row of pipettes 36. When one or more tips 62 are present within the row of wells 63 registered with the sensor, the light beam 82 from the LED will be broken and will not reach the photoelectric element 90. However, if all of the tips in a row are successfully picked up by the pipette assembly, the beam will extend across the tray 56 and be detected by the photoelectric element. By proper positioning LED 89 and photoelectric element 90, possible pick up of tray 56 itself, as by friction between tips 62 and wells in tray 56, can also be detected.

Figs. 12 to 14 illustrate an alternative embodiment of the moveable table arrangement of the present

invention. This embodiment includes another microtiter tray 88 between sample tray 54 and tip supply tray 56. Tray 88 may contain either a liquid supply of biological material or a reagent for initially filling the titer tray receptacles. While shown as a plurality of individual wells, tray 88 may be a common supply trough or pan. For example the initial charge of sample material may be injected into a first row of receptacles in tray 54 and after replacement of tips 62 from tray 56, the remaining wells in 56 filled with diluent transferred from another portion of tray 88 or a separate supply of liquid from another tray. As mentioned above, after use, pipette tips may be ejected into empty receptacles in tip tray 56. However, it is also contemplated that another portion of table 10 would permit collection of all used tips. This may either be a slot in table 10 (not shown) permitting drop of the tips into a bin below the table, or a collection bin located at another position on the table.

In operation, the automatic serial dilutor basically functions to pick up a row of tips in the tray 56, insert them in one row of wells in the titer tray 54, extract some of the liquid sample from these wells, inject the tips into the diluent in the next successive row of wells, oscillate the plungers to mix the liquid, position the tips to expel all liquid and then return the tips to the tray 56. This operation is set forth in greater detail with reference to the following example of a program that can be used by the microprocessor to effect a serial dilution process.

Step	Command	Action
001	Table to position M	Bring row M of tray 56 under pipettes
002	Head assembly down	Load tips
003	Head assembly up	Pick up tips
004	Detect for complete tip pick-up	Yes: Go to 005 No: go to 002
005	Table to position N	Bring row N of tray 54 under tips
006	Head assembly down	Insert tips in wells
007	Plunger up	Aspirate sample into pipettes
008	Head assembly up	Remove tips from wells
009	Table to position N + 1	Bring next row of tray 54 under tips
010	Head assembly down	Insert tips in wells
011	Oscillate plunger	Mix sample and diluent
012	Head assembly up partially	Tips just out of liquid in wells
013	Plunger down	Expel sample from pipette
014	Head assembly slightly further	Above meniscus
015	Plunger down beyond initial point	Expel all of sample and some air
016	Head assembly to top position	
017	Plunger up to initial point	
018	Table to position M	Bring empty row of tray 56 under tips
019	Head down	Insert tips in receptacles
020	Tip ejector down	Release tips

Step	Command	Action
021	Tip ejector up	
022	Detect for complete Tip ejection	Yes: Go to 023 No: Go to 020
023	Head assembly up	
024	M = M + 1, N = N + 1	
025	Table to position M	
026	Go to 002	

The cycle is repeated a number of times equal to the number of dilutions to be carried out. During any given cycle steps 001—004 and 18—22 can be deleted if changing of the tips is not required.

Prior to the initiation of a serial dilution operation, the microprocessor 70 can be programmed with the volume of liquid that is to be transferred during each cycle of the process. This amount determines the extent to which the plunger rods 40 are raised during step 007 of the program. This action, in turn, determines the concentration of the sample in successive wells of the tray 54. For example, to obtain a dilution spectrum in which the concentration in one row is one-half that of the preceding row, the first row of wells might be filled with 100)1 of the sample and all other wells filled with 50)1 of diluent each. The microprocessor would be set up to cause 50)1 to be transferred from one well to the next succeeding well during each cycle.

During step 011, the plunger rods 40 can be oscillated up and down about 5 times to assure adequate mixing.

At the beginning of each cycle of the serial dilution process, the plunger rods 40 are disposed at a predetermined calibration point within the pipettes. A Hall-effect sensor similar to the type described previously with respect to the table 10 can be used to monitor and control the position of the rods. In step number 014 of the program, after the sample and diluent have been mixed in step 011, the plunger tips are raised so that they are just above the level of liquid in the wells in step 012 and the plunger is returned to the calibration point to expel the liquid from the pipettes in step 013, the tips are raised to a point just above the meniscus of liquid in the receptacle. By then extending the plungers downwardly beyond the calibration point, all liquid is expelled from the pipettes. This action effectively blows the liquid out of the pipettes by causing some air trapped within the pipette to also be ejected and permits any liquid remaining in the tips and extending between the tip and the receptacle to be drawn out of the tip by capillary action due to surface tension acting on the liquid. This step is particularly effective where the tip is made of a non-wettable plastic which as above noted is a preferred material.

Although certain steps have been shown to be discrete, they can be executed simultaneously. For example, steps 016 and 017 might take place at the same time.

It will be appreciated by those of ordinary skill in the art that the present invention can be embodied in other specific forms without departing from the spirit or essential characteristics thereof. The presently disclosed embodiment is therefore considered in all respects to be illustrative and not restrictive. For example, where the term "stepper" motor has been used to describe the preferred embodiment of the motor drive means for table, head assembly and plunger mechanism, it will be apparent that other precise positioning means may be used, such as direct current servo motors. The scope of the invention accordingly is indicated by the appended claims.

Claims

1. An apparatus for handling small liquid samples during filling, transferring or mixing of such liquid samples by repetitive use of the same pipette (36) to transfer liquids between a plurality of sample receptacles (63) having different quantities or types of liquid therein which includes a plurality of pipettes (36), each having a piston section (39) and a pipette tip member (62) securable thereto for insertion into said sample receptacles (63) to withdraw liquids from or inject liquids into said receptacles (63), and a mounting block (37) having a cylinder (35) formed therein for housing the piston section (39), a plunger (40) positioned in each said pipette (36) for recriprocal motion through said pipette (36), wherein a fluid seal (47, 49) is arranged between the plunger (40) and the cylinder (35),

an elongated pipette tip member (62) for covering one end of said piston section (39) and forming a substantial portion of the fluid volume of said pipette (36), said tip member (62) having one end frictionally engageable with a portion of the sidewall surface of said piston section (39) to form a fluid seal with it, characterized by

reciprocal means (64) having a member slidably surrounding the exterior of said piston section to contact said tip member (62) to frictionally disengage said tip member (62) from said sidewall surface,

means (56, 63) for supporting a plurality of said pipette tip members (62) in a vertical position for selective frictional engagement with said sidewall surface,

means (30, 32) for reciprocating each of said piston sections (39) to frictionally engage said tip members (62) with said surface,

means (66, 68) for actuating said reciprocal means (64) to disengage said tip member (62) from said piston section (39), and

an actuating means (28) for actuating the reciprocating means (30, 32).

2. Apparatus in accordance with claim 1 wherein each of said pipettes is resiliently mounted to said apparatus to permit independent, relative movement between each said piston section (39).

3. Apparatus in accordance with claim 1 or 2 wherein said plunger moving means includes a stepper motor (46) and a lead screw drive mechanism (48) interconnecting said stepper motor (46) and said plungers (40).

4. Apparatus in accordance with any of claims 1 to 3, wherein the plurality of the pipettes (36) are arranged in a row and form a plunger assembly (34), the plunger assembly (34), the reciprocal means (64) for disengaging tip members (62) from said sidewall surfaces, the means (66, 68) for actuating said reciprocal means (64) and the means (30, 32) for reciprocating each of the piston section (39) are supported by a head assembly (12), the head assembly (12) is translatable between upper and lower positions along a vertical axis and means (28) are provided for moving said head assembly along said vertical axis.

5. Apparatus in accordance with any of claims 1 to 4 further comprising means (18, 20, 22) for horizontal movement of the table (10).

6. Apparatus in accordance with claim 5 wherein the means for horizontal movement comprises a stepper motor (18), a pinion (20) and a rack (22), which are mounted on the underside of the table (10).

7. Apparatus according to any of claims 1 to 6, further comprising controlling means (70) for controlling the operation of each actuating means for movement of the pipettes (36), said plunger (40), said reciprocal means (64) and said table (10).

8. Apparatus according to any of claims 1 to 7, further comprising sensor means (72, 74) for detecting the position of the table (10).

9. Apparatus according to claim 8, further comprising limit sensors (76) which are disposed at the respective ends of the path of travel of the table (10).

10. Apparatus according to any of claims 1 to 9, further comprising sensor means (89, 90) for detecting the presence of tip members (62) in said sample receptacles (63).

11. Apparatus according to any of claims 6 to 10, characterized in that the controlling means (70) monitors the movements by controlling signals of said sensor means.

12. A method of operation of a computer to control a liquid handling device having a plurality of pipettes with electrically operable plungers to alter their internal volumes for aspiration and dispensing of liquid, removable tips and an electrically operable tip ejection mechanism and transport means including a table driven by a mechanism under the control of said computer for horizontally moving liquid receptacles in a first work station and including a head assembly driven by a mechanism under the control of said computer for transporting the pipettes so that the disposable tips may be placed in a plurality of liquid receptacles arranged on said table, at least some of said liquid receptacles having liquid therein and having at least one row of said disposable tips stored in a second work station on said table for automatic transfer of liquid samples between said plurality of liquid receptacles, the method comprising the steps of:

a) causing the computer to run a program which causes said computer to issue the proper electrical commands to said electrically operable plungers of said pipettes, said driving mechanisms for said table and head assembly of said transport means and to said tip ejection mechanism to cause the following movements to occur in sequence;

b) moving said table to register a row of pipette tips stored on said second work station with the pipettes on said head assembly;

c) lowering said head assembly and said pipettes to engage a row of said disposable tips in the registered row and raise said head assembly to pick up the engaged tips with said pipettes;

d) moving said table to index at least some of said liquid containing receptacles at said first work station under said row of pipettes;

e) moving said head assembly so as to insert the engaged tips in the liquid in said liquid containing receptacles and moving said plunger so as to withdraw at least some of the liquid therefrom into said pipettes;

f) raising said head assembly and pipettes;

g) moving said table to index one or more other liquid receptacles under said pipettes;

h) lowering said head assembly and pipettes and moving said plungers so as to expel the liquid in said pipettes into the other receptacles;

i) raising said head assembly and pipettes;

j) moving the table to bring a selected position at said second work station into alignment with said pipettes;

k) move head down and move tip ejection mechanism so as to discharge said engaged tips from said pipettes;

l) raise the head assembly.

13. The method of claim 12 which includes repeating the steps for a set of unused tips and a new set of liquid receptacles until a desired number of transfers or dilutions are completed.

14. The method of claim 12 or 13 wherein the step of expelling liquid into said receptacle includes at least once after expelling liquid into said receptacle, moving said plunger so as to aspirate liquid from said receptacle into said pipettes and again moving said plunger so as to expel liquid from said pipettes to effect mixing of the contents of said receptacle with the added liquid.

15. The method of any of claims 12 to 14 wherein expelling of liquids in said pipettes in step (h) includes actuation of a plunger in each pipette while said tips are just above the meniscus of liquid in said receptacles to expel any remaining liquid in said tips through capillary attraction between liquid in said tips and said receptacle.

16. The method of any of claims 12 to 15 further including the step of detecting whether all of the tips in a row are successfully picked up during step (c).

17. The method of claim 12 wherein said tips are discharged onto said second work station during step (k).

18. The method of claim 12 which includes the further step of detecting whether all tips have been ejected after step (k).

19. The method of any of claims 12 to 18 wherein steps d) through i) are repeated before steps j) through l) are performed.

20. The method of any of claims 12 to 18 which includes repeating steps b) through l) for successive rows of tips and successive receptacles at said respective work stations until a desired number of transfers or dilutions are completed.

Patentansprüche

1. Vorrichtung zum Handhaben von kleinen Flüssigkeitsproben beim Füllen, Übertragen oder Mischen solcher Flüssigkeitsproben durch wiederholte Verwendung der gleichen Pipette (36) zum Übertragen von Flüssigkeiten zwischen mehreren Probenbehältern (63) mit darin befindlichen

unterschiedlichen Mengen oder Arten von Flüssigkeiten, mit mehreren Pipetten (36), die jeweils einen Kolbenabschnitt (39) und ein Pipettenspitzenelement (62) aufweisen, das zum Einführen in die Probenbehälter (63) daran befestigbar ist, so daß Flüssigkeiten entweder aus den Behältern (63) entnommen oder in diese eingespritzt werden, und mit einem Befestigungsblock (37) mit einem darin ausgebildeten Zylinder (35) zur Aufnahme des Kolbenabschnitts (39), einem in der Pipette (36) angeordneten Kolben (40), der durch die Pipette (36) hin- und herbewegbar ist, wobei eine Flüssigkeitsdichtung (47, 49) zwischen dem Kolben (40) und dem Zylinder (35) angeordnet ist,

wobei das längsgestreckte Pipetten-Spitzenelement (62) ein Ende des Kolbenabschnitts (39) abdeckt und einen wesentlichen Abschnitt des Flüssigkeitsvolumens der Pipette (36) bildet, wobei das Spitzenelement (62) ein Ende aufweist, das reibungsschlüssig mit einem Abschnitt der Seitenwandfläche des Kolbenabschnitts (39) in Eingriff bringbar ist und so eine Flüssigkeitsdichtung mit dieser gebildet wird, gekennzeichnet durch

eine hin- und hergehende Einrichtung (64) mit einem Element, das das Äußere des Kolbenabschnitts gleitend umgibt zum Berühren des Spitzenelements (62) und Lösen des Reibungseingriffs des Spitzenelements (62) von der Seitenwandfläche,

eine Einrichtung (56, 63) zum Tragen mehrerer Pipetten-Spitzenelemente (62) in einer senkrechten Stellung zum wahlweisen reibungsschlüssigen Ineingriffnehmen mit der Seitenwandfläche,

eine Einrichtung (30, 32) zum Hin- und Herbewegen jedes der Kolbenabschnitte (39), so daß die Spitzenelemente (62) reibungsschlüssig mit der genannten Fläche in Eingriff kommen,

eine Einrichtung (66, 68) zum Betätigen der sich hin- und herbewegenden Einrichtung (64) zum Lösen des Spitzenelements (62) von dem Kolbenabschnitt (39) und

eine Betätigungseinrichtung (28) zum Betätigen der sich hin- und herbewegenden Einrichtung (30, 32).

2. Vorrichtung nach Anspruch 1, wobei jede der Pipetten federnd an der Vorrichtung befestigt ist, wodurch eine unabhängige, relative Bewegung zwischen jedem Kolbenabschnitt (39) möglich ist.

3. Vorrichtung nach Anspruch 1 oder 2, wobei die Kolbenbetätigungseinrichtung einen Schrittmotor (46) und eine den Schrittmotor (46) und die Kolben (40) verbindende Führungsschrauben-Antriebsmechanik (48) aufweist.

4. Vorrichtung nach einem der Ansprüche 1 bis 3, wobei die mehreren Pipetten (36) in einer Reihe angeordnet sind und eine Kolbenanordnung (34) bilden, wobei die Kolbenanordnung (34), die sich hin- und herbewegende Einrichtung (64) zum Lösen der Spitzenelemente (62) von den Seitenwandflächen, die Einrichtung (66, 68) zum Betätigen der sich hin- und herbewegenden Einrichtung (64) und die Einrichtung (30, 32) zum Hin- und Herbewegen jedes Kolbenabschnitts (39) von einer Kopfanordnung (12) getragen werden, wobei die Kopfanordnung (12) zwischen oberen und unteren Stellungen entlang einer vertikalen Achse verstellbar ist und Einrichtungen (28) zum Bewegen der Kopfanordnung entlang der vertikalen Achse vorgesehen sind.

5. Vorrichtung nach einem der Ansprüche 1 bis 4, ferner mit einer Einrichtung (18, 20, 22) für eine horizontale Bewegung eines Tisches (10).

6. Vorrichtung nach Anspruch 5, wobei die Einrichtung für die Horizontalbewegung einen Schrittmotor

(18), ein Ritzel (20) und eine Zahnstange (22) aufweist, die an der Unterseite des Tisches (10) befestigt sind.

7. Vorrichtung nach einem der Ansprüche 1 bis 6, ferner mit einer Steuereinrichtung (70) zum Steuern des Betriebs wie jeder Betätigungseinrichtung für die Bewegung der Pipetten (36), des Kolbens (40), der sich hin- und herbewegenden Einrichtung (64) und des Tisches (10).

8. Vorrichtung nach einem der Ansprüche 1 bis 7, ferner mit Sensoreinrichtungen (72, 74) zum Detektieren der Position des Tisches (10).

9. Vorrichtung nach Anspruch 8, ferner mit Grenzsensoren (76), die an den jeweiligen Enden des Bewegungswegs des Tisches (10) angeordnet sind.

10. Vorrichtung nach einem der Ansprüche 1 bis 9, ferner mit Sensoreinrichtungen (89, 90) zum Detektieren des Vorhandenseins von Spitzenelementen (62) in den Probenbehältern (63).

11. Vorrichtung nach einem der Ansprüche 6 bis 10, dadurch gekennzeichnet, daß die Steuereinrichtung (70) die Bewegungen überwacht mit Hilfe von Steuersignalen der Sensoreinrichtungen.

12. Verfahren zum Betrieb eines Computers zum Steuern einer Flüssigkeits-Handhabungsvorrichtung mit mehreren Pipetten, die elektrisch betätigbare Kolben aufweist zum Verändern des inneren Volumens zum Aufnehmen und Abgeben von Flüssigkeit, abnehmbaren Spitzen und einer elektrisch betätigbaren Spitzenabstoßmechanik und mit einer Transporteinrichtung, die einen durch eine computergesteuerte Mechanik angetriebenen Tisch für eine horizontale Bewegung von Flüssigkeitsbehältern in einer ersten Arbeitsstation aufweist und eine durch eine computergesteuerte Mechanik angetriebene Kopfanordnung zum Transportieren der Pipetten aufweist, so daß die Wegwerfspitzen in mehrere in dem Tisch angeordnete Flüssigkeitsbehälter angeordnet werden können, wobei mindestens einige der Flüssigkeitsbehälter Flüssigkeit enthalten und wobei mindestens eine Reihe der Wegwerfspitzen in einer zweiten Arbeitsstation gelagert sind zum automatischen Übertragen von Flüssigkeitsproben zwischen den mehreren Flüssigkeitsbehältern, wobei das Verfahren folgende Schritte aufweist:

a) Bewirken, daß der Computer ein Programm durchläuft, wodurch der Computer die richtigen elektrischen Befehle an die elektrisch betätigbaren Kolben der Pipetten, an die Antriebsmechaniken für den Tisch und die Kopfanordnung der Transporteinrichtung und an die Spitzenausstoßmechanik liefert, wodurch die folgenden Bewegungen in Folge veranlaßt werden;

b) Bewegen des Tisches, so daß eine Reihe von in der zweiten Arbeitsstation gelagerten Pipettenspitzen mit den Pipetten an der Kopfanordnung ausgerichtet sind,

c) Absenken der Kopfanordnung und der Pipetten zum Ineingriffbringen einer Reihe der Wegwerfspitzen in der ausgerichteten Reihe und Anheben der Kopfanordnung zum Aufnehmen der in Eingriff gebrachten Spitzen mit den Pipetten,

d) Bewegen des Tisches, so daß mindestens einige der Flüssigkeit enthaltenden Behälter an der ersten Arbeitsstation unter der Pipettenreihe zu liegen kommen,

e) Bewegen der Kopfanordnung, so daß die in Eingriff gebrachten Spitzen in die Flüssigkeit der Flüssigkeit enthaltenden Behälter eingeführt werden und Bewegen des Kolbens, so daß mindestens ein Teil der Flüssigkeit daraus in die Pipetten entnommen wird,

f) Anheben der Kopfanordnung und der Pipetten,

g) Bewegen des Tisches, so daß ein oder mehrere andere Flüssigkeitsbehälter unter den Pipetten zu liegen kommen,

h) Absenken der Kopfanordnung und der Pipetten und Bewegen der Kolben, so daß die Flüssigkeit in den Pipetten in die anderen Behälter ausgestoßen wird,

i) Anheben der Kopfanordnung und der Pipetten,

j) Bewegen des Tisches, so daß eine ausgewählte Position an der zweiten Arbeitsstation mit den Pipetten ausgerichtet wird,

k) Absenken des Kopfes und Bewegen der Spitzenausstoßmechanik, so daß die in Eingriff gebrachten Spitzen von den Pipetten abgeworfen werden,

l) Anheben der Kopfanordnung.

13. Verfahren nach Anspruch 12, wobei die Schritte für einen Satz ungebrauchter Spitzen und einen neuen Satz von Flüssigkeitsbehältern wiederholt werden, bis eine gewünschte Anzahl von Übertragungen oder Verdünnungen abgeschlossen ist.

14. Verfahren nach Anspruch 12 oder 13, wobei der Schritt beim Ausstoßen von Flüssigkeit in den Behälter nach dem Ausstoßen von Flüssigkeit in den Behälter mindestens einmal den folgenden Vorgang aufweist, Bewegen des Kolbens, so daß Flüssigkeit aus dem Behälter in die Pipetten aufgenommen wird und erneut das Bewegen des Kolbens, so daß die Flüssigkeit aus den Pipetten ausgestoßen wird, so daß ein Mischen der Inhalte der Behälter mit den zugefügten Flüssigkeiten bewirkt wird.

15. Verfahren nach einem der Ansprüche 12 bis 14, wobei das Ausstoßen von Flüssigkeiten in den Pipetten im Schritt (h) umfaßt Betätigen eines Kolbens in jeder Pipette, während die Spitzen kurz oberhalb des Meniskus der Flüssigkeit in den Behältern sind, so daß etwaige Flüssigkeitsreste in den Spitzen durch Kapillaranziehungskraft zwischen der Flüssigkeit in den Spitzen und den Behältern ausgestoßen wird.

16. Verfahren nach einem der Ansprüche 12 bis 15 mit einem weiteren Schritt zum Detektieren, ob alle Spitzen in einer Reihe während des Schritts (c) erfolgreich aufgenommen wurden.

17. Verfahren nach Anspruch 12, wobei die Spitzen während des Schritts (k) auf die zweite Arbeitsstation abgeladen werden.

18. Verfahren nach Anspruch 12, mit dem weiteren Schritt zum Detektieren, ob alle Spitzen nach dem Schritt (k) abgestoßen wurden.

19. Verfahren nach einem der Ansprüche 12 bis 18, wobei die Schritte d) bis i) wiederholt werden, bevor die Schritte j) bis l) ausgeführt werden.

20. Verfahren nach einem der Ansprüche 12 bis 18, wobei die Schritte b) bis l) für aufeinanderfolgende Reihen mit Spitzen und aufeinanderfolgende Behälter an zugehörigen Arbeitsstationen wiederholt werden, bis eine bestimmte Anzahl von Übertragungen oder Verdünnungen abgeschlossen ist.

Revendications

1. Appareil pour la manipulation de petits échantillons liquides pendant le remplissage, le transfert ou le mélange de ces échantillons liquides par utilisation répétitive de la même pipette (36) pour transférer des liquides entre plusieurs réceptacles à échantillons (63) contenant différentes quantités ou différents types de liquides, qui comprend plusieurs pipettes (36) comportant chacune une section de piston (39) et un embout de pipette (62) pouvant y être fixé en vue d'une introduction dans les réceptacles à échantillons (63) pour soutirer des liquides des réceptacles (63) ou y injecter des liquides, et un bloc de montage (37) dans lequel est formé un cylindre (35) destiné à loger la section de piston (39) ainsi qu'un plongeur (40) placé dans chaque pipette (36) afin de se déplacer en va-et-vient à travers la pipette (36), un joint d'étanchéité au fluide (47, 49) étant prévu entre le plongeur (40) et le cylindre (35),

un long embout de pipette (62) pour couvrir une extrémité de la section de piston (39) et former une partie substantielle du volume de fluide de la pipette (36), l'embout (62) comportant une extrémité pouvant être engagée à friction avec une partie de la surface de paroi latérale de la section de piston (39) pour former un joint étanche au fluide avec celle-ci,

caractérisé par

un dispositif à va-et-vient (64) comportant un élément entourant à coulissement l'extérieur de la section de piston afin de venir en contact avec l'embout (62) pour dégager cet embout (62) par friction de la surface de paroi latérale,

des moyens (56, 63) pour supporter plusieurs embouts de pipettes (62) dans une position verticale en vue d'un engagement à friction sélectif avec la surface de paroi latérale,

un dispositif (30, 32) pour animer chacune des sections de pistons (39) d'un mouvement de va-et-vient afin d'engager les embouts (62) par friction avec la surface,

des moyens (66, 68) pour actionner le dispositif à va-et-vient (64) afin de dégager l'embout (62) de la section de piston (39), et

un dispositif d'actionnement (28) pour actionner le dispositif (30, 32).

2. Appareil suivant la revendication 1, dans lequel chacune des pipettes est montée élastiquement sur l'appareil afin de permettre un déplacement relatif indépendant entre les sections de pistons (39).

3. Appareil suivant la revendication 1 ou 2, dans lequel le dispositif servant à déplacer les plongeurs comprend un moteur pas à pas (46) et un mécanisme d'entraînement à vis mère (48) reliant le moteur pas à pas (46) et les plongeurs (40).

4. Appareil suivant l'une quelconque des revendications 1 à 3, dans lequel les pipettes (36) sont disposées en une rangée et forment un ensemble de plongeurs (34), l'ensemble de plongeurs (34), le dispositif à va-et-vient (64) pour dégager les embouts (62) des surfaces des parois latérales, les moyens (66, 68) pour actionner le dispositif à va-et-vient (64) et le dispositif (30, 32) pour animer chaque section de piston (39) d'un mouvement de va-et-vient sont supportés par une tête (12), la tête (12) peut être déplacée entre des positions supérieure et inférieure suivant un axe vertical, et des moyens (28) sont prévus pour déplacer la tête le long de l'axe vertical.

5. Appareil suivant l'une quelconque des revendications 1 à 4 comprenant, en outre, un dispositif (18, 20, 22) pour assurer le déplacement horizontal de la table (10).

6. Appareil suivant la revendication 5, dans lequel le dispositif pour assurer le déplacement horizontal comprend un moteur pas à pas (18), un pignon (20) et une crémaillère (22) qui sont montés sur le dessous de la table (10).

7. Appareil suivant l'une quelconque des revendications 1 à 6 comprenant, en outre, un dispositif de commande (70) pour commander le fonctionnement de chaque dispositif d'actionnement en vue du déplacement des pipettes (36), du plongeur (40), du dispositif à va-et-vient (64) et de la table (10).

8. Appareil suivant l'une quelconque des revendications 1 à 7 comprenant, en outre, des capteurs (72, 74) pour détecter la position de la table (10).

9. Appareil suivant la revendication 8 comprenant, en outre, des capteurs de fin de course (76) qui sont disposés aux extrémités respectives du trajet de la table (10).

10. Appareil suivant l'une quelconque des revendications 1 à 9 comprenant, en outre, des capteurs (89, 90) pour détecter la présence d'embouts (62) dans les réceptacles à échantillons (63).

11. Appareil suivant l'une quelconque des revendications 6 à 10, caractérisé en ce que le dispositif de commande (70) surveille les déplacements en contrôlant les signaux des capteurs.

12. Procédé de mise en oeuvre d'un ordinateur pour commander un dispositif manipulateur de liquide comportant plusieurs pipettes avec des plongeurs actionnés électriquement pour modifier leurs volumes intérieurs afin d'aspirer et de débiter du liquide, des embouts amovibles et un mécanisme d'éjection

d'embouts actionné électriquement, ainsi qu'un dispositif de transport comprenant une table entraînée par un mécanisme sous la commande de l'ordinateur pour déplacer horizontalement des réceptacles à liquide dans un premier poste de travail et comprenant une tête entraînée par un mécanisme sous la commande de l'ordinateur pour transporter les pipettes de telle façon que les embouts jetables puissent être placés dans plusieurs réceptacles à liquide disposés sur la table, au moins certains réceptacles à liquide contenant du liquide et comportant une rangée d'embouts jetables stockés dans un second poste de travail sur la table afin de transférer automatiquement des échantillons liquides entre les divers réceptacles à liquide, ce procédé comprenant les opérations suivantes:

a) l'ordinateur exécute un programme qui le force à émettre les ordres électriques adéquats vers les plongeurs de pipettes actionnés électriquement, les mécanismes d'entraînement pour la table et la tête du dispositif de transport et le mécanisme d'éjection d'embouts afin d'amener les déplacements suivants à se dérouler dans l'ordre;

b) la table est déplacée pour amener une rangée d'embouts de pipettes stockés sur le second poste de travail en coïncidence avec les pipettes prévues sur la tête;

c) la tête et les pipettes sont abaissées pour engager une rangée des embouts jetables constituant la rangée coïncidente et la tête est élevée pour saisir les embouts engagés en même temps que les pipettes;

d) la table est déplacée pour amener au moins certains des réceptacles contenant du liquide au premier poste de travail sous la rangée de pipettes;

e) la tête est déplacée de manière à introduire les embouts engagés dans le liquide présent dans les réceptacles contenant du liquide et les plongeurs sont déplacés de manière à soutirer au moins une fraction du liquide dans les pipettes;

f) la tête et les pipettes sont déplacées vers le haut;

g) la table est déplacée pour amener un ou plusieurs autres réceptacles à liquide en dessous des pipettes;

h) la tête et les pipettes sont abaissées et les plongeurs sont déplacés de manière à expulser le liquide contenu dans les pipettes dans les autres réceptacles;

i) la tête et les pipettes sont déplacées vers le haut;

j) la table est déplacée pour amener une position choisie au second poste de travail en ligne avec les pipettes;

k) la tête est abaissée et le mécanisme d'éjection des embouts est déplacé de manière à décharger les embouts engagés des pipettes, et

l) la tête est déplacée vers le haut.

13. Procédé suivant la revendication 12, suivant lequel les opérations sont répétées pour un jeu d'embouts non utilisés et pour un nouveau jeu de réceptacles à liquide jusqu'à ce qu'un nombre souhaité de transferts ou de dilutions aient été effectués.

14. Procédé suivant la revendication 12 ou 13, dans lequel l'opération consistant à expulser du liquide dans le réceptacle comprend au moins, après avoir expulsé du liquide dans le récipient, un déplacement du plongeur en vue d'aspirer du liquide du réceptacle dans les pipettes et un nouveau déplacement du plongeur de manière à expulser du liquide des pipettes en vue de mélanger le liquide présent dans les réceptacles avec le liquide ajouté.

15. Procédé suivant l'une quelconque des revendications 12 à 14, dans lequel l'expulsion de liquides dans des pipettes dans l'opération h) comprend l'actionnement d'un plongeur dans chaque pipette, tandis que les embouts se trouvent juste au-dessus du ménisque liquide présent dans les réceptacles afin d'expulser tout liquide résiduel dans les embouts par attraction capillaire entre le liquide dans les embouts et le réceptacle.

16. Procédé suivant l'une quelconque des revendications 12 à 15 comprenant, en outre, l'opération qui consiste à détecter si tous les embouts d'une rangée sont effectivement saisis pendant l'opération c).

17. Procédé suivant la revendication 12, dans lequel les embouts sont déchargés sur le second poste de travail pendant l'opération k).

18. Procédé suivant la revendication 12, qui comprend l'opération supplémentaire consistant à détecter si tous les embouts ont été éjectés après l'opération k).

19. Procédé suivant l'une quelconque des revendications 12 à 18, dans lequel les opérations d) à i) sont répétées avant que les opérations j) à l) soient exécutées.

20. Procédé suivant l'une quelconque des revendications 12 à 18, qui comprend la répétition des opérations d) à l) pour des rangées successives d'embouts et des réceptacles successifs aux postes de travail respectifs jusqu'à ce qu'un nombre souhaité de transferts ou de dilutions aient été effectués.

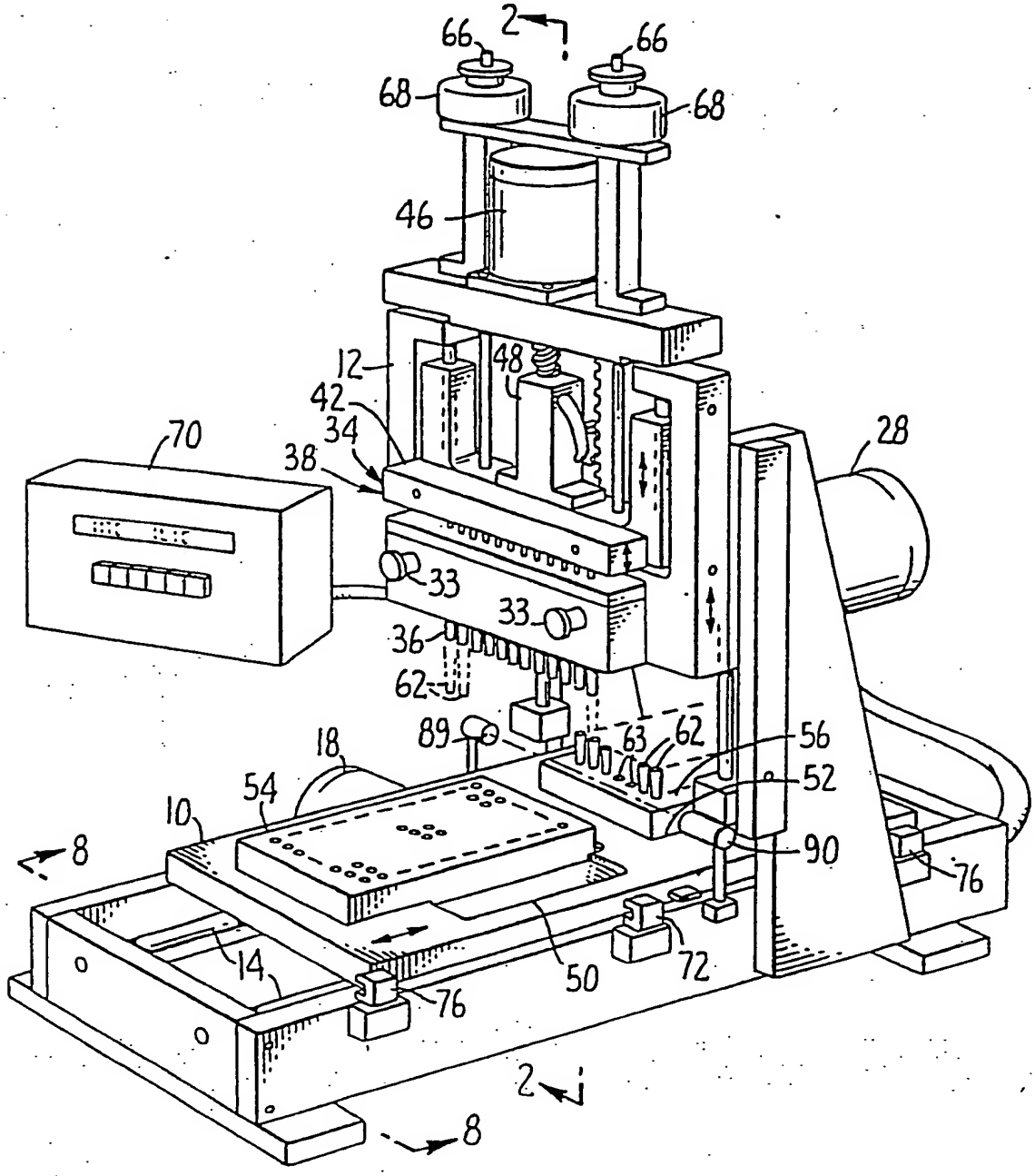
66
2
66
68
68
46
12
48
42
34
38
70
28
33
33
36
62
89
63
62
56
52
18
54
10
8
90
76
14
50
72
76
2
8

FIG. 1.

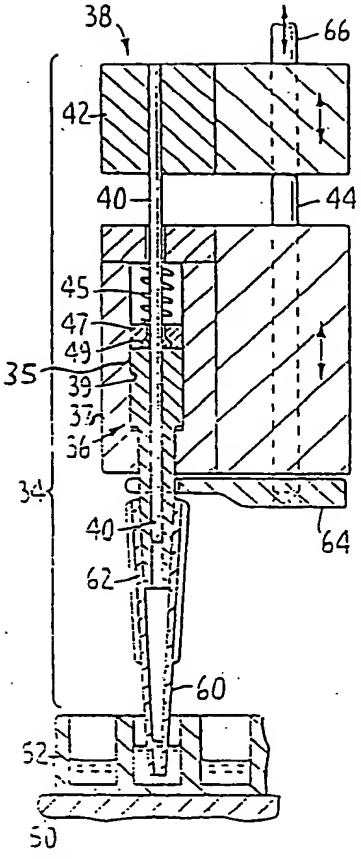

FIG. 3.

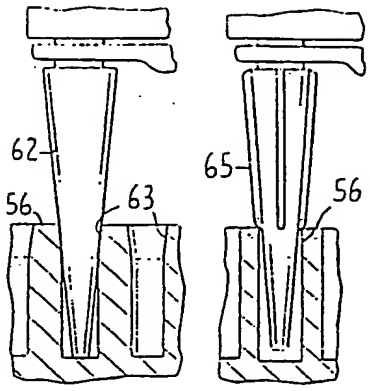

FIG. 4.

FIG. 5.

FIG. 2.

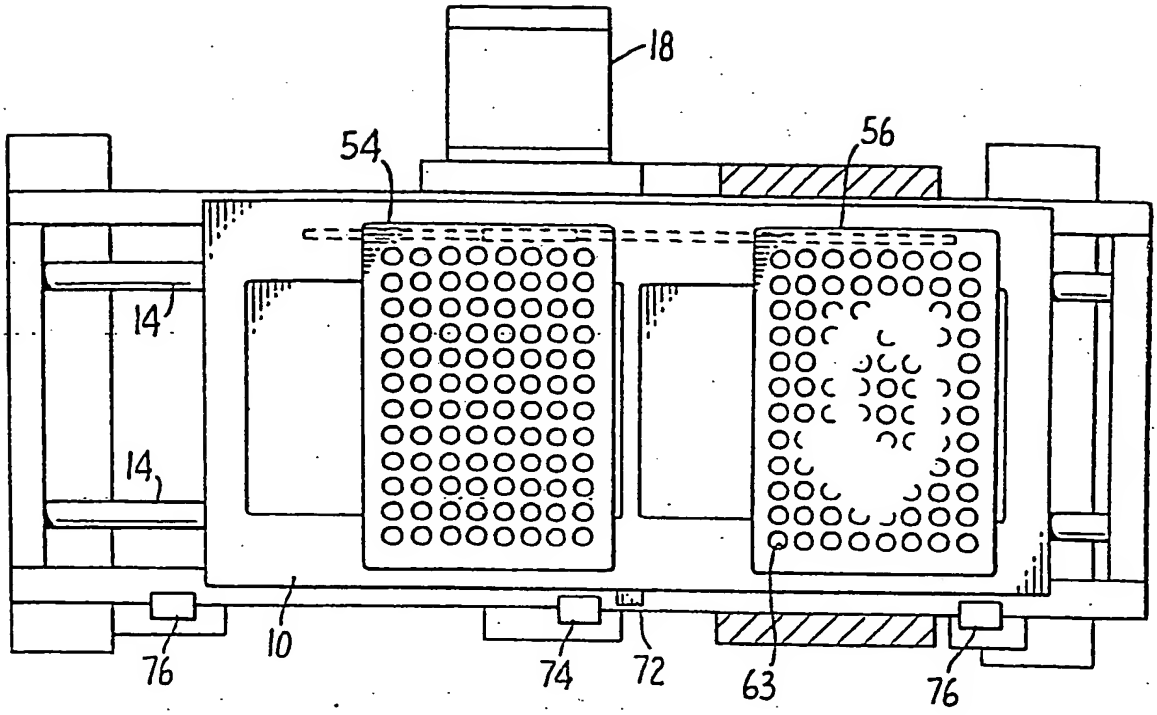

FIG. 6.

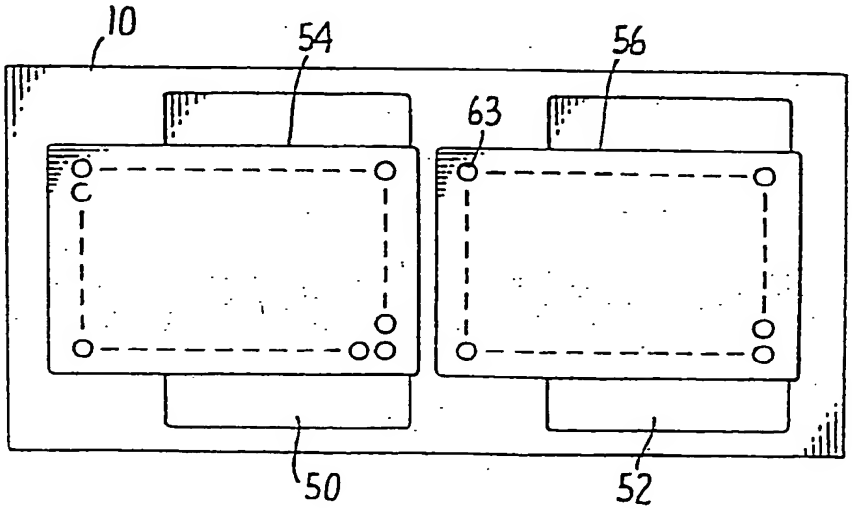

FIG. 7.

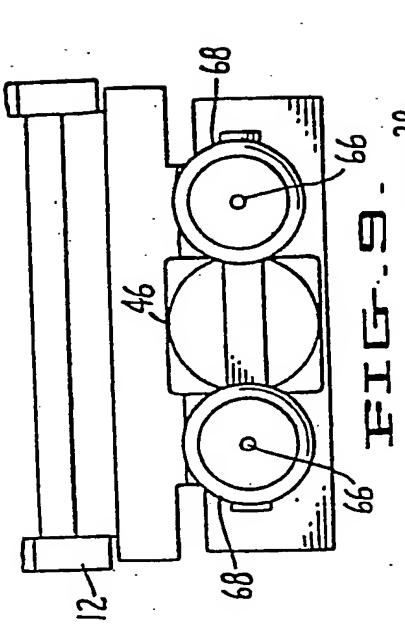

FIG-9-

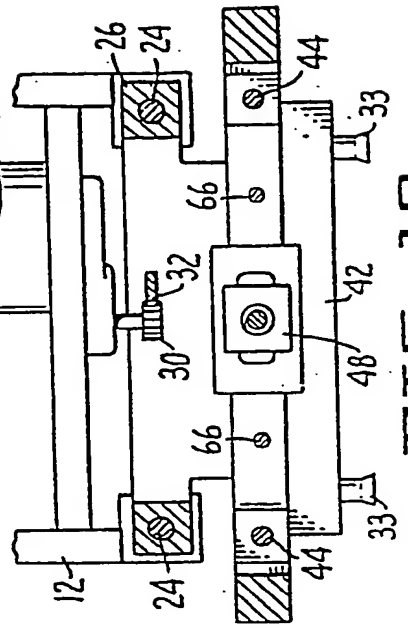

FIG-10-

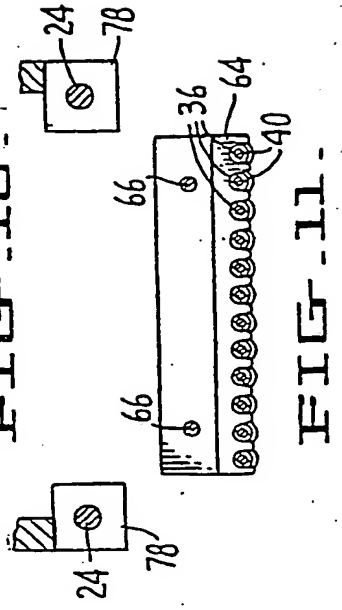

FIG-11-

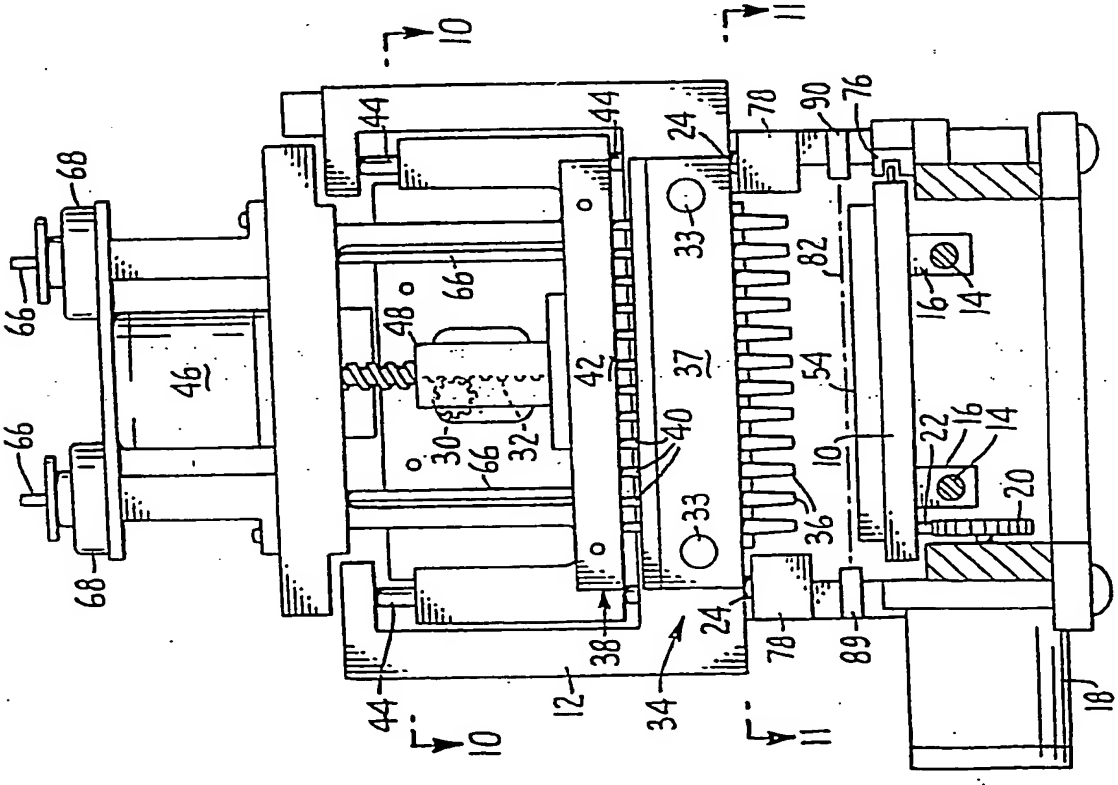

FIG-8-

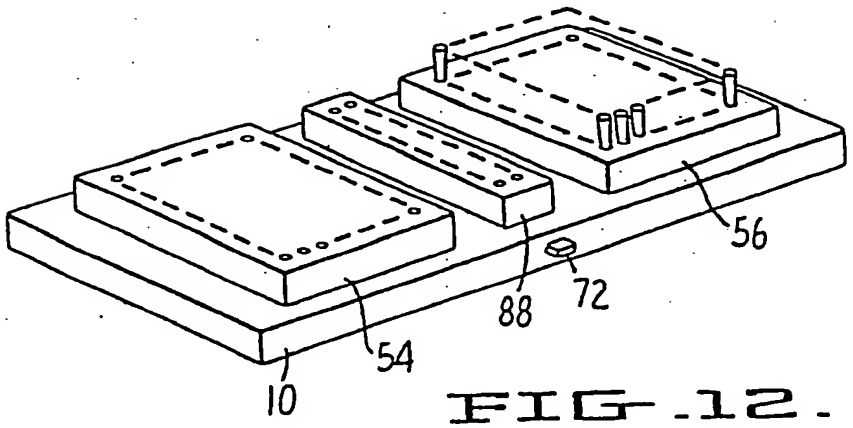

FIG. 12.

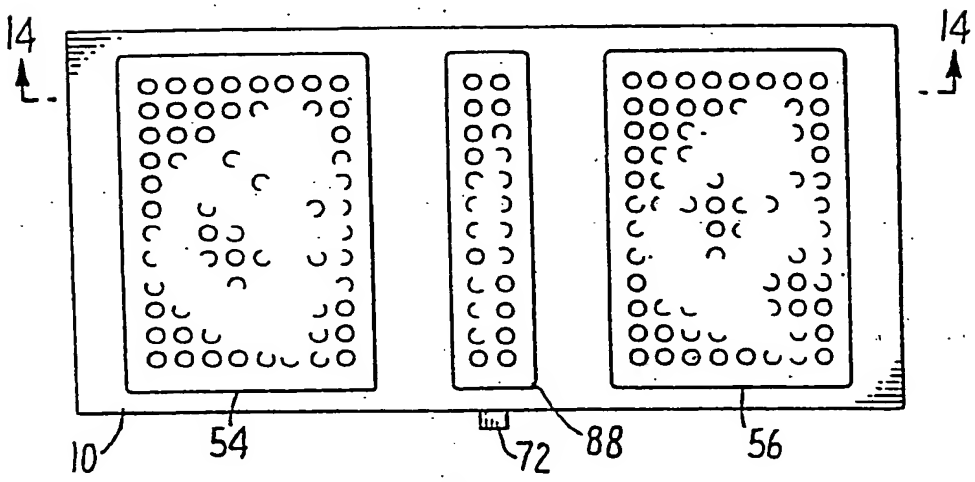

FIG. 13.

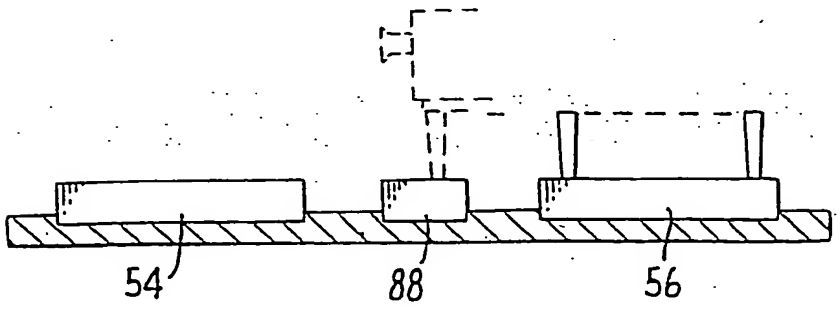

FIG. 14.